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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
10/699,394	10/31/2003	Henriette Draborg	10308.200-US	6155

25908 7590 06/28/2007

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EXAMINER	
MOORE, WILLIAM W	

ART UNIT	PAPER NUMBER
1656	

MAIL DATE	DELIVERY MODE
06/28/2007	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/699,394	DRABORG ET AL.	
	Examiner	Art Unit	
	William W. Moore	1656	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *18 January 2007*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *16-55* is/are pending in the application.

4a) Of the above claim(s) *49 and 55* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *16-48 and 50-5455* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

Continued Examination Under 37 CFR 1.114

A request for continued examination under 37 CFR 1.114, including the fee set forth in 37 CFR 1.17(e), was filed in this application after final rejection. Since this application is eligible for continued examination under 37 CFR 1.114, and the fee set forth in 37 CFR 1.17(e) has been timely paid, the finality of the previous Office action has been withdrawn pursuant to 37 CFR 1.114. Applicant's submission filed on 5 December 2006 has been entered.

Response to Amendment

Applicant's Amendments to claims 16, 20, 22, 32, 47, and 48 in the Response filed 25 December 2006 overcome the rejections of record of claims herein under the second paragraph of 35 U.S.C. § 112 as well as the rejections of record of claims herein under 35 U.S.C. § 102. Applicant's telephonic election of the invention of the protease products, and compositions that comprise them, of the original claims 1-9 was commemorated at page 3 of the communication mailed 29 September 2005, together with the further election of a protease species comprising any naturally-occurring amino acid, or a relative insertion or deletion of an amino acid, at the subtilisin BPN'-correspondent position 62, together with a further modification that is either a N76D or an N76S substitution. Applicant affirmed the election at page 17 of the Response filed 29 March 2006. Claims 16-54 include the elected species but claim 49, and the new claim 55, are withdrawn from consideration as drawn to a non-elected invention because claim 49 requires a ten-fold substitution but the only such substitution set disclosed in the specification, at page 11 at the bottom of the right hand column, lacks a substitution at the position of the elected species and because the new claim 55 excludes an amino acid substitution at the position of the elected species, i.e., modification at the subtilisin BPN'-correspondent position 62.

The obviousness-type double patenting rejection of record of claims herein over claim 1 of US Patent No. 6,245,901 is withdrawn because alignment of SEQ ID NO:2 of the patent and SEQ ID NO:1 herein indicates that the subtilisin BPN'-correspondent position 62 in the PD948 protease amino acid sequence is not the position 62 recited in the patent claim, where arginine resides and is replaced by lysine, but is instead position 70 of SEQ ID NO:2 of the patent, where an asparagine occurs. Asparagine also occurs at position 62 of the subtilisin BPN' sequence of SEQ ID NO:1 herein. The provisional obviousness-type double patenting rejection of record of claims herein over the copending, commonly-assigned, Application No. 09/957,806 is withdrawn because the claims 1-21 remaining in the copending application are all drawn to methods of selection. This rejection is not made final because new grounds of rejection are stated below

where several references applied as prior art to the elected species of primary substitution, i.e., a generic substitution at the subtilisin BPN'-correspondent position 62, can also be applied as prior art to other positions for generic amino acid substitutions recited in the preamble of claim 16, thus retiring the use of the reference for this set of claims.

Claim Objections

Claims 16 and 46 are objected to because of the following informalities: Claims 16 and 46 erroneously recite "**N296K**", a position to which no position in the 275-amino acid sequence of the mature subtilisin BPN corresponds. This is clearly an error, as these claims should instead recite the substitution **N269K**. See page 12, line 3, of the specification. Appropriate correction is required to correct the error, e.g., amending the claims to delete N296K and state "N269T,K".

Double Patenting

The nonstatutory double patenting rejection is based on a judicially created doctrine grounded in public policy (a policy reflected in the statute) so as to prevent the unjustified or improper timewise extension of the "right to exclude" granted by a patent and to prevent possible harassment by multiple assignees. See *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985); *In re Van Ornum*, 686 F.2d 937, 214 USPQ 761 (CCPA 1982); *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970); and *In re Thorington,* 418 F.2d 528, 163 USPQ 644 (CCPA 1969).

A timely filed terminal disclaimer in compliance with 37 CFR 1.321(c) may be used to overcome an actual or provisional rejection based on a nonstatutory double patenting ground provided the conflicting application or patent is shown to be commonly owned with this application. See 37 CFR 1.130(b). Effective January 1, 1994, a registered attorney or agent of record may sign a terminal disclaimer. A terminal disclaimer signed by the assignee must fully comply with 37 CFR 3.73(b).

Claims 16-18, 22, 25, 34-36, 39, 46, 53, and 54 herein are provisionally rejected under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 34-36 and 39-55 of U.S. Patent No. 6,245,901. Although the conflicting claim are not identical, they are not patentably distinct from each other because protease modifications of the claims 34-36 and 39-53 of the copending application, and detergent compositions comprising same of claims 54 and 55 of the copending application include – based on generic substitutions permitted at subtilisin BPN'-correspondent positions 68 and 131 according to claim 16 herein – the paired modifications and sets of modifications of subtilases of claims 16-18, 22, 25, 34-36, 39, 46, herein, in particular the set indicated in claim 48 herein, and the detergent compositions comprising same of claims 53 and 54 herein.

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of 35 U.S.C. § 102 that form the basis for the rejections under this section made in this Office action:

A person shall be entitled to a patent unless –

(b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

(e) the invention was described in a patent granted on an application for patent by another filed in the United States before the invention thereof by the applicant for patent, or on an international application by another who has fulfilled the requirements of paragraphs (1), (2), and (4) of section 371(c) of this title before the invention thereof by the applicant for patent.

Claims 16, 21, 22, 27, 31, 34, 39, 43, and 50-54 are rejected under 35 U.S.C. § 102(b) as being anticipated by Åaslyng et al. US 5,665,587, made of record herewith.

Åaslyng et al. disclose protease variants comprising the substitution sets G97D+H120K, S99D+N140K, R170Y+G195E, R170Y+K251E, R170Y+G195E+K251E, and P14D+T22K+ . . . N76D+A98R+S99D+H120D+N140D in both a sub-group I-S1 subtilisin, subtilisin Carlsberg, and a sub-group I-S2 subtilisin, subtilisin 309, where the indicated positions are numbered according to the amino acid sequence of subtilisin BPN' set forth in SEQ ID NO:1 herein, meeting the limitations of claims 16, 21, 22, 27, 31, 34, 39, 43, and 50-52 herein. See cols. 10-13, 19-21 and 33. Åaslyng et al. also disclose preparation of detergent compositions comprising these protease variants and a surfactant as well as an amylase or a lipase, meeting limitations of claims 53 and 54 herein. See cols. 22-24 and 35-40.

Claims 16-18, 26, 29, 30, 32, 35, 36, 39-41 and 51-54 are rejected under 35 U.S.C. § 102(b) as being anticipated by Brode et al. US 6,599,730, made of record in the communication mailed 29 September 2005.

Brode et al. '730 disclose subtilisin 309 variants, i.e., variants of the amino acid sequence set forth in SEQ ID NO:2 herein which is a sub-group I-S2 subtilisin, that comprise the following substitution sets, where subtilisin 309 amino acid sequence positions are indicated in brackets, and their numbering by correspondence with the amino acid sequence of subtilisin BPN' is indicated in parentheses and where Applicant's convention of a comma separating substituents is replaced by a diagonal mark separating the several substituent amino acids that meet the limitations of claims 16-18, 26, 29, 30, 32, 35, 36, 39-41 and 51 herein:

N(62)[60]S/Q/D/E + G(61)[59]S/E/D,
N(62)[60]S/Q/D/E + G(97)[95]P/S/N/Q/D/E,
N(62)[60]S/Q/D/E + A(98)[96]H/P/S/T/G/N/Q/D/E,
N(62)[60]S/Q/D/E + S(99)[97]D/E,
N(62)[60]S/Q/D/E + G(102)[100]S/D,
N(62)[60]S/Q/D/E + S(106)[104]D/E,
N(62)[60]S/Q/D/E + I(107)[105]/V/M/T,
N(62)[60]S/Q/D/E + L(126)[124]I,
N(62)[60]S/Q/D/E + G(127)[125]E,

N(62)[60]S/Q/D/E + P(131)[129]N/Q/S/N/D/E,
N(62)[60]S/Q/D/E + A(158)[156]T//D/E,
N(62)[60]S/Q/D/E + Y(167)[161]A/I,
N(62)[60]S/Q/D/E + V(203)[197]L/M/A/I/Q,
N(62)[60]S/Q/D/E + N(204)[198]D/S,
N(62)[60]S/Q/D/E + Y(209)[203]C/H,
N(62)[60]S/Q/D/E + G(211)[205]D,
N(62)[60]S/Q/D/E + Y(214)[208]C/H, and
N(62)[60]S/Q/D/E + N(218)[212]S/D.

See, e.g., col. 8, line 38, through col. 11, line 44. Brode et al. disclose that subtilisin variants comprising these amino acid substitutions will provide an improved wash performance when formulated in a detergent composition because they will have "decreased absorption to, and increased hydrolysis of, an insoluble substrate" when employed in a method of cleaning textiles or surfaces and accordingly disclose the preparation of detergent compositions comprising these protease variants and a surfactant and further comprising, e.g., cellulases, lipases, and amylases, meeting the limitations of claims 53 and 54. See the abstract and cols. 97-106.

Claims 16-22, 24, 26, 30, 32, 34, 37-39, 41-46 and 50-54 are rejected under 35 U.S.C. § 102(e) as being anticipated by Ghosh et al. US 6,376,450, made of record herewith.

Ghosh et al. disclose multiply-substituted subtilisins wherein amino acid substitutions at the subtilisin BPN'-correspondent position 62 are combined with the amino acid substitutions G20R, N76D, N185D, and S256R, where these positions occur in any subtilisin and are also numbered by correspondence with the amino acid sequence of subtilisin BPN'. See Table II at cols. 17-30. Ghosh et al. also disclose multiply-substituted subtilisins wherein amino acid substitutions at the subtilisin BPN'-correspondent position 68 are combined with the amino acid substitutions S3L, I8V, G20R, N18S, G20A, T22K, K27R, N43D, P55S, G61E, N76D, A98T, S99D, A114V, N116D, N116S, N173D, A174V, N183D, N184S, N184D, N185D, V203A, N204D, Q206L, N218D, T224A, A230V, N252K, Q245R, Q245L, S256R, S256N, T260R, T260A, and N261D, where these positions occur in any subtilisin and are also numbered by correspondence with the amino acid sequence of subtilisin BPN'. See Table II at cols. 17-30.

Ghosh et al. also disclose multiply-substituted subtilisins wherein amino acid substitutions at the subtilisin BPN'-correspondent position 97 are combined with the amino acid substitutions Q245R, and N252K where these positions occur in any subtilisin and are also numbered by correspondence with the amino acid sequence of subtilisin BPN'. See Table II at cols. 17-30. Ghosh et al. further disclose multiply-substituted subtilisins wherein amino acid substitutions at the subtilisin BPN'-correspondent position 98 are combined with the amino acid substitutions

V68A, N76D, Q245R, and N252K, where these positions occur in any subtilisin and are also numbered by correspondence with the amino acid sequence of subtilisin BPN'. See Table II at cols. 17-30. Ghosh et al. further disclose as well multiply-substituted subtilisins wherein amino acid substitutions at the subtilisin BPN'-correspondent position 99 are combined with the amino acid substitutions V68A, N76D, N184D, N204T, Q245R, and N252K, where these positions occur in any subtilisin and are also numbered by correspondence with the amino acid sequence of subtilisin BPN'. See Table II at cols. 17-30. Ghosh et al. additionally disclose multiply-substituted subtilisins wherein an amino acid substitution at the subtilisin BPN'-correspondent position 170 are combined with the amino acid substitutions V68A, N76D, S130T, N185D, N185S, M222S, and Q245R where these positions occur in any subtilisin and are also numbered by correspondence with the amino acid sequence of subtilisin BPN'. See Table II at cols. 17-30.

Ghosh et al. also disclose multiply-substituted subtilisins wherein amino acid substitutions at the subtilisin BPN'-correspondent position 245 are combined with the amino acid substitutions S3L, I8V, N18S, G20A, G20R, K27R, N43D, S57P, G61E, N62D, N76D, V68A, G97E, A98D, A98L, A98T, A98V, S99E, S99G, S99N, A114V, N116D, N116S, S130T, N140D, R170S, N173D, A174V, N183D, N184D, N184S, N185D, N185S, V203A, N204D, Q206L, N218D, N218S, M222S, T224A, A230V, V244A, V244I, K251R, N252K, N252L, N252F, T255S, S256R, S256N, S259G, T260A, T260R, N261D, and L262S, where these positions occur in any subtilisin and are also numbered by correspondence with the amino acid sequence of subtilisin BPN'. See Table II at cols. 17-30. Ghosh et al. then disclose multiply-substituted subtilisins wherein amino acid substitutions at the subtilisin BPN'-correspondent position 252 are combined with amino acid substitutions S3L, V4E, I8V, N18S, G20A, G20R, N43D, V68A, G61E, N62D, N76D, G97E, A98D, A98L, A98V, S99E, S99G, S99N, N116S, N140D, N173D, N184D, N184S, N185D, N218S, Q245R, S256R, S256N, T260R, and N261D, where these positions occur in any subtilisin and are also numbered by correspondence with the amino acid sequence of subtilisin BPN'. See Table II at cols. 17-30.

Ghosh et al. additionally disclose that subtilisin variants comprising combinations of their many sets of amino acid substitutions may be formulated into cleaning compositions, including detergent compositions, to "provid[e] improved and enhanced cleaning of fabrics, dishware, tableware, kitchenware, cookware, and other hard surface substrates". See col. 4, lines 45-53, and col. 65, line 1, through col. 108, line 50.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. § 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

Claims 16-18, 27, 48, 50-54 are rejected under 35 U.S.C. § 103(a) as obvious over any one of Ballinger et al. US 5,741,664, US 5,780,285, and US 5,837,516, all made of record herewith, and Gosselink et al. US 6,121,226, made of record herewith.

Ballinger et al. teach the modification of the sub-group I-S1 subtilisin BPN' having the amino acid sequence set forth in SEQ ID NO:1 herein by replacing an asparagine at position 62 with aspartate or glutamate to alter its native specificity for hydrophobic, or small, uncharged, amino acids, providing variants with a higher degree of specificity for cleavage at basic amino acid residues. See, e.g., col. 2, line 66, to col. 3, line 16 and claims 1 and 2 of Ballinger et al. '664. While Gosselink et al. do not specifically teach the preparation of variant subtilisins they teach the preparation of detergent compositions comprising a surfactant and any of several enzymes, such as amylases, lipases, cellulases, and peroxidases, and comprise as well variant subtilisins having an amino acid substitution that replaces any amino acid present in any subtilisin at a position corresponding to position 62 of the mature subtilisin BPN', a class I-S1 subtilase having the amino acid sequence of SEQ ID NO:1 herein, with aspartate. Gosselink et al. further teach that amino acid substitutions in either a sub-group I-S1 subtilase, such as subtilisin BPN', or a sub-group I-S2 subtilase, such as subtilisin 309, may be combined when made at positions that include the subtilisin BPN'-correspondent positions 1, **3**, **4**, **8**, 9, **10**, 13, 16, 17, **18**, 19, **20**, 21, **22**, 24, **27**, 38, **43**, **48**, 55, **57**, **61**, **62, 68**, 72, 75, **76**, 77, 78, 87, 89, 101, **102**, **107**, 111, **114**, **116**, 119, 121, **126**, 128, **130**, 133, 134, 137, **140**, **158**, 160, **167**, **174, 183, 184, 185**, 188, 192, **203**, **204**, **209**, **211**, 212, 213, **214**, **218**, **222, 224**, 228, **230**, **237**, 238, 240, 242, **244, 251**, 254, **255, 256,** 257, 258, **259, 260, 261, 262,** 263, **265**, 268, 269, 271, and 274, among which an amino acid substitution replaces any amino acid present at the subtilisin BPN' position 76 in any subtilisin with aspartate. Moreover, Gosselink et al. specifically teach the nine-fold substitution set of claim 48 herein. See cols. 27-31, particularly col. 29 at lines 38-67 and col. 30 at line 31 through col. 31 at line 4.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to make a subtilisin variant of claims 16-18 and 27 herein by combining either of the

N62D or N62E modifications taught by Ballinger et al. with the modification X76D taught by Gosselink et al. to be a common prior art modification made in either a sub-group I-S1 subtilase, such as subtilisin BPN', or a sub-group I-S2 subtilase, such as subtilisin 309, and to prepare a detergent composition comprising such a variant subtilisin of claims 53 and 54 herein. This is because both Ballinger et al. and Gosselink et al. teach that it is advantageous to make an amino acid substitution at the BPN'-correspondent position 62 in a subtilisin, because asparagine is present at the subtilisin BPN'-correspondent position 62 in both sub-group I-S1 subtilases and sub-group I-S2 subtilases thus such an artisan at that time would have had a reasonable expectation of success in modifying the substrate specificity of either a sub-group I-S1 subtilase or a sub-group I-S2 subtilase by making an N62D or an N62E subtilisin, and because Gosselink et al. teach that the prior art of subtilisin modification shows that a substitution of X76D, an N76D substitution in subtilisin BPN' as well as at the corresponding position in subtilisin 309, was generally regarded to be advantageously combined with further amino acid substitutions in modifying a subtilisin to make it more effective for use in detergent compositions. It would have also have been obvious to one of ordinary skill in the art at the time the invention was made to make subtilisin variants of claim 48 herein by combining either the N62D substitution or the N62E substitution taught by Ballinger et al. with the prior art nine-fold substitution set because Gosselink et al. teach that the nine-fold substitution set stated in claim 48 herein is commonly regarded as a beneficial set of substitutions to combine with another advantageous amino acid substitutions in modifying a subtilisin amino acid sequence.

Claims 16-18, 21, 22, 23, 26, 27, 31, 32, 34, 38, 41, 43, 46, 47, and 50-54 are rejected under 35 U.S.C. § 103(a) as obvious over Ballinger et al. and Gosselink et al. as applied to claims 16-18, 27, 48, 50-54 above, further in view of Aaslyng et al. US 5,665,587 and Brode et al. US 6,599,730, both previously cited.

The teachings of Ballinger et al., Gosselink et al., Brode et al., and Aaslyng et al., all discussed above, are take as before, particularly the teachings of Brode et al. of combining the amino acid substitutions of N62S/Q/D/E with further substitutions at as many as eighteen other positions in the amino acid sequence of subtilisin 309, set forth in SEQ ID NO:2, herein, including the subtilisin BPN'-correspondent positions 97, 98, 99, 158, 167, 209, 214 and 218. Like Gosselink et al., Aaslyng et al. teach the preparation of variant subtilisins comprising a replacement of the amino acid at the subtilisin BPN'-correspondent position 76 with aspartate where the amino acid sequence may be that of a sub-group I-S1 subtilase, including the subtilisins BPN', DY and Carlsberg, or that of a sub-group I-S2 subtilases, including subtilisins 309 and 147 and the PB92 protease, and specifically teach the combination of the N76D substitution with further amino acid substitutions, each at positions identified by correspondence

with the amino acid sequence of the mature subtilisin BPN' provided in SEQ ID NO:1 herein, including P14D, T22K, K27R, G97D, S99D, A98R, H120D, N140D, and G195E. See cols 19-21. Aaslyng et al. also teach the preparation of variant subtilisins that comprise the amino acid substitutions, each at positions identified by correspondence with the amino acid sequence of the mature subtilisin BPN' provided in SEQ ID NO:1 herein, P14K, V51D, P129D, H120K, N140K, N185D, N218D, N218S, K237R, K251E, K251R, and S265R. See also cols. 19-21, col. 42 at line 47, and claim 1. Aaslyng et al. teach that each of these substitutions, whether made singly or in sets of substitutions and whether the result of the substitution is achieved in either a sub-group I-S1 subtilase or a sub-group I-S2 subtilase, provides a variant subtilisin advantageous for formulation in detergent compositions because the variant will have an isoelectric point shifted to achieve an optimum wash performance by matching the pH of the wash liquor comprising the detergent composition wherein the variant "is intended for use". See col. 18 al lines 6-16.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to combine the N62D substitution of both Ballinger et al. and Brode et al. with the N76D substitution of both Brode et al. and Aaslyng et al. and to further combine the pair of amino acid substitutions with one or more of the further amino acid substitutions of Brode et al., set forth in the rejection above under 35 U.S.C. § 102(b), as well as with one or more of the further amino acid substitutions P14D, P14K, T22K, K27R, V51D, G97D, S99D, A98R, H120D, H120K, P129D, N140D, N140K, N185D, N218D, N218S, K237R, K251E, K251R, and S265R taught by Aaslyng et al., including the three-fold substitution set of claim 47, N26D+Y214H+K237R, and to prepare a detergent composition comprising such a variant subtilisin and a surfactant of claims 53 and 54 herein. This is because Brode et al., Ballinger et al., and Gosselink et al. teach the advantages of introducing an amino acid substitution at the BPN'-correspondent position 62 in a subtilisin, such as the N62D substitution of Brode et al. where both sub-group I-S1 subtilases and sub-group I-S2 subtilases have asparagine present at the subtilisin BPN'-correspondent position 62, because Gosselink et al. teach that the prior art of subtilisin modification shows that a substitution of X76D, an N76D substitution in the subtilisin BPN' amino acid sequence and the corresponding position in the subtilisin 309 amino acid sequence, was well known to be advantageously combined with further amino acid substitutions in modifying a subtilisin to make it more effective for use in detergent compositions, and because Aaslyng et al. teach that their net molecular charge-altering substitutions, whether made individually or in sets of substitutions, will shift the isoelectric point of a variant subtilisin that comprises one or more of them in order to achieve an optimum wash performance of variant at any target pH of the wash liquor wherein its

use is intended. Such an artisan would have had a reasonable expectation of success in preparing a subtilisin variant having an improved wash performance at a target pH of wash liquor wherein its use is intended and an improved wash performance in the presence of insoluble substrates when combining a substitution of Brode et al. at the subtilisin BPN'-correspondent position 62 with an N76D substitution of Brode et al. and Aaslyng et al. and a further substitution of one or more of the P14D, P14K, T22K, K27R, V51D, H120D, P129D, N140K, N185D, N218D, N218S, K237R, K251E, K251R, and S265R substitutions of Aaslyng et al. because Aaslyng et al. themselves combine the N76D substitution with several further substitutions and because Brode et al. teach that their N62D substitution will improve the wash performance of a variant subtilisin by providing a decreased absorption to, and increased hydrolysis of, insoluble substrates when combined with further substitutions.

Claims 16-19, 21, 25-27, 30, 31, 33, 34, 42, 46 and 51-54 are rejected under 35 U.S.C. § 103(a) as obvious over Ballinger et al. and Gosselink et al. as applied to claims 16-18, 27, 48, 50-54 above, further in view of both Brode et al., previously cited, and Christianson et al., US 5,340,735, made of record herewith.

The teachings of Ballinger et al., Gosselink et al., and Brode et al., discussed above, are taken as before. Christianson et al. teach that introducing amino acid substitutions in the amino acid sequence of a mature subtilisin will increase its shelf stability and retain its catalytic activity in detergent compositions by structural stabilization of the resulting subtilisin variant, exemplified with a sub-group I-S2 subtilisin, primarily by enhancing van der Waals interactions in the interior regions. See col. 1, at lines 40-56, and col. 19, line 22, through col. 20, line 24. In view of their definitions of "small, hydrophobic amino acid" and "small amino acid" at lines 45-56 of col. 8, Christianson et al. teach the preparation of subtilisin variants that comprise the following substitutions, where parentheses indicate the subtilisin BPN'-correspondent position, brackets indicate the subtilisin BLAP amino acid sequence position, and diagonal marks separate the substituent amino acids indicated in claims 16, 21, 25, 26, 30, 31, 33, 34, 42 and 46 herein:

S(3)[3]T/A;
V(4)[4]A/C;
A(48)[47]T;
T(71)[69]A;
S(106)[104]T;
N(116)[114]S;
H(120)[118]N/D/Q/K/E/Y/S;
S(132)[130]T/G;
T(143)[141]A;

A(230)[224]V; and
T(274)[268]A.

See particularly, col. 8, line 49, through col. 10, line 4.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to combine the N62D substitution taught by Ballinger et al. and Brode et al. with the N76D substitution of both Brode et al. and Aaslyng et al. and to further combine the pair of amino acid substitutions with one or more of the S3T, V4A/C, A48T, T71A, S106T, N116S, S132T/G, H120N/D/Q/K/E/Y/S, T143A, A230V and T274A amino acid substitutions taught by Christianson et al. in any sub-group I-S2 subtilisin of claim 51 herein, including the subtilisin 309 of claim 52 herein, and to prepare a detergent composition comprising such a variant subtilisin and a surfactant of claims 53 and 54 herein. This is because Brode et al., Ballinger et al., and Gosselink et al. teach the advantages of introducing an amino acid substitution at the BPN'-correspondent position 62 in a subtilisin, such as the N62D substitution of Brode et al. where both of sub-groups I-S1 and I-S2 subtilases have asparagine present at the subtilisin BPN'-correspondent position 62, because Gosselink et al. teach that the prior art of subtilisin modification shows that a substitution of X76D, an N76D substitution in the subtilisin BPN' amino acid sequence and the corresponding position in the subtilisin 309 amino acid sequence, was well known to be advantageously combined with further amino acid substitutions in modifying a subtilisin to make it more effective for use in detergent compositions, and because Christianson et al. teach that each of their stabilizing amino acid substitutions will provide a variant subtilisin that has an increased shelf stability and retains its catalytic activity in detergent compositions. It would also have been obvious to such an artisan at that time to prepare a detergent composition that comprised such a variant sub-group I-S2 subtilisin as well as cellulases, lipases, and amylases, because Christianson et al. teach that their substitutions meet a need for an improved, variant, subtilisin used in such compositions and because Ballinger et al., Gosselink et al., and Brode et al. teach that it is advantageous to incorporate such further enzymes in detergent compositions. Such an artisan would have had a reasonable expectation of success in preparing a subtilisin variant having improved wash performance as well as an increased shelf stability and retained catalytic activity in detergent compositions when combining an N62S/Q/D substitution and an N76D substitution of Brode et al. with any of the S3T, V4A/C, A48T, T71A, N116S, H120N/D/Q/K/E/Y/S, S132T/G, T143A, A230V and T274A of Christianson et al. because the amino acid substitutions of Brode et al. are all at solvent-exposed positions of a subtilisin and the amino acid substitutions of Christianson et al. are primarily made at positions at the internal surfaces of a sub-group I-S2 subtilase.

Claims 16-19, 26, 27, 41 and 51-54 are rejected under 35 U.S.C. § 103(a) as obvious over Ballinger et al. and Gosselink et al. as applied to claims 16-18, 27, 48, 50-54 above, further in view of both Brode et al., previously cited, and Sierkstra et al., US 5,837,517, made of record herewith.

The teachings of Ballinger et al., Gosselink et al., and Brode et al., discussed above, are taken as before. Sierkstra et al. teach that introducing one or more of the substitutions S57P, N76D, H120D, G195E, N218S, M222A, and M222S in the amino acid sequence of subtilisin 309, where these substitutions are numbered by correspondence with the amino acid sequence of subtilisin BPN', will increase its wash performance and/or stability in a detergent composition. See the abstract and the patent claims. It would have been obvious to one of ordinary skill in the art at the time the invention was made to combine the N62D substitution taught by Ballinger et al. and Brode et al. with the N76D substitution of both Brode et al. and Sierkstra et al. and to further combine one or more of the S57P, H120D, G195E, N218S, M222A, and M222S amino acid substitutions taught by Sierkstra et al. in any sub-group I-S2 subtilisin of claim 51 herein, including the subtilisin 309 of claim 52 herein, and to prepare a detergent composition comprising such a variant subtilisin and a surfactant of claims 53 and 54 herein. This is because Brode et al., Ballinger et al., and Gosselink et al. teach the advantages of introducing an amino acid substitution at the BPN'-correspondent position 62 in a subtilisin, such as the N62D substitution of Brode et al. where both of sub-groups I-S1 and I-S2 subtilases have asparagine present at the subtilisin BPN'-correspondent position 62, because Sierkstra et al. explicitly teach that an N76D substitution in the subtilisin 309 amino acid sequence is advantageously combined with further amino acid substitutions in modifying a subtilisin 309 to make it more effective for use in detergent compositions, and because Sierkstra et al. teach that each of further their stabilizing amino acid substitutions S57P, H120D, G195E, N218S, M222A, and M222S will provide a variant subtilisin 309 that has an increased wash performance and/or stability in a detergent composition. It would also have been obvious to such an artisan at that time to prepare a detergent composition that comprised such a variant subtilisin 30 as well as cellulases, lipases, and amylases, because Sierkstra et al. teach that their substitutions meet a need for an improved, variant, subtilisin used in such compositions and because Ballinger et al., Gosselink et al., and Brode et al. teach that it is advantageous to incorporate such further enzymes in detergent compositions. Such an artisan would have had a reasonable expectation of success in preparing a subtilisin 309 variant having improved wash performance as well as an increased wash performance and stability in detergent compositions when combining an N62S/Q/D/E substitution of Brode et al. and an N76D substitution of Brode et al. and Sierkstra et al. with any of the further S57P, H120D, G195E, N218S, M222A, and M222S substitutions of

Sierkstra et al. because the N62S/Q/D/E amino acid substitutions of Brode et al. are compatible with those made Sierkstra et al. and also improve the wash performance of a subtilisin 309 variant.

Claims 19-22, 24, 26, 30, 32, 34, 37-39, 41-46, and 50-54 are rejected under 35 U.S.C. § 103(a) as obvious over Ballinger et al. and Gosselink et al. as applied to claims 16-18, 27, 48, 50-54 above, further in view of both Brode et al. and Ghosh et al., both discussed above.

The teachings of Ballinger et al., Gosselink et al., Brode et al., and Ghosh et al., discussed above, are taken as before, particularly the teaching of Ghosh et al. of the combination of the amino acid substitution N76D with the amino acid substitutions S3L, I8V, N18S, G20A, G20R, T22K, K27R, N43D, P55S, S57P, G61E, N62D, N76D, V68A, G97E, A98D, A98L, A98T, A98V, S99D, S99E, S99G, S99N, A114V, N116D, N116S, S130T, N140D, R170S, N173D, A174V, N183D, N184D, N184S, N185D, N185S, V203A, N204D, Q206L, N218D, N218S, M222S, T224A, A230V, V244A, V244I, N252K, Q245R, Q245L, K251R, N252K, N252L, N252F, T255S, S256R, S256N, S259G, T260A, T260R, N261D, and L262S. It would have been obvious to one of ordinary skill in the art at the time the invention was made to combine the N62D substitution taught by Ballinger et al. and Brode et al. with the N76D substitution of both Brode et al. and Ghosh et al. and to further combine the pair of amino acid substitutions with one or more of the S3L, I8V, N18S, G20A, G20R, T22K, K27R, N43D, P55S, S57P, G61E, N62D, N76D, V68A, G97E, A98D, A98L, A98T, A98V, S99D, S99E, S99G, S99N, A114V, N116D, N116S, S130T, N140D, R170S, N173D, A174V, N183D, N184D, N184S, N185D, N185S, V203A, N204D, Q206L, N218D, N218S, M222S, T224A, A230V, V244A, V244I, N252K, Q245R, Q245L, K251R, N252K, N252L, N252F, T255S, S256R, S256N, S259G, T260A, T260R, N261D, and L262S amino acid substitutions taught by Ghosh et al. in a subtilisin of claims 50 and 51 herein, as well as the subtilisin 309 of claim 52 herein, and to prepare a detergent composition comprising such a variant subtilisin and a surfactant of claims 53 and 54 herein. This is because Brode et al., Ballinger et al., and Gosselink et al. teach the advantages of introducing an amino acid substitution at the BPN'-correspondent position 62 in a subtilisin, such as the N62D substitution of Brode et al. where both of sub-groups I-S1 and I-S2 subtilases have asparagine present at the subtilisin BPN'-correspondent position 62, because Gosselink et al. teach that the prior art of subtilisin modification shows that a substitution of X76D, an N76D substitution in the subtilisin BPN' amino acid sequence and the corresponding position in the subtilisin 309 amino acid sequence, was well known to be advantageously combined with further amino acid substitutions in modifying a subtilisin to make it more effective for use in detergent compositions, and because Ghosh et al. teach that their sets of amino acid

substitutions, including both N62D and N76D substitutions will provide subtilisin variants having improved and enhanced cleaning of fabrics, dishware, tableware, kitchenware, cookware, and other hard surface substrates. It would also have been obvious to such an artisan at that time to prepare a detergent composition that comprised such a variant sub-group I-S2 subtilisin as well as cellulases, lipases, and amylases, because Ghosh et al. teach that their substitutions meet a need for an improved, variant, subtilisin used in such compositions and because Ballinger et al., Gosselink et al., and Brode et al. teach that it is advantageous to incorporate such variant subtilisins in detergent compositions. Such an artisan would have had a reasonable expectation of success in preparing a subtilisin variant having improved wash performance as well as an increased shelf stability and retained catalytic activity in detergent compositions when combining an N62S/Q/D substitution and an N76D substitution of Brode et al. with any of the substitutions of Ghosh et al. because both Brode et al. and Ghosh et al. teach that their amino acid substitutions provide improved wash performance in the cleaning of both fabrics and hard surface substrates.

Claims 28, 50, 51, 53 and 54 are rejected under 35 U.S.C. § 103(a) as obvious over Ballinger et al. and Gosselink et al. as applied to claims 16-18, 27, 48, 50-54 above, further in view of both Brode et al., previously cited, and Bott et al., US 5,700,676, made of record herewith.

The teachings of Ballinger et al., Gosselink et al., and Brode et al., discussed above, are taken as before. Bott et al. teach the preparation of the amino acid substitution S87C in the amino acid sequence of subtilisin BPN', a class I-S1 subtilisin in order to increase both its thermal stability and resistance to autocatalytic cleavage by the formation of a disulfide bond between the positions 24 and 87. See Example 11 at cols. 67-70, where this substitution pair provides improved stability and nearly equivalent autocatalytic cleavage characteristics by comparison with the native, or wild-type, subtilisin BPN'. It would have been obvious to one of ordinary skill in the art at the time the invention was made to combine the N62D substitution taught by Ballinger et al. and Brode et al. with the N76D substitution of Brode et al. and to further combine the pair of amino acid substitutions that includes the S87C substitution taught by Bott et al. in at least a sub-group I-S1 subtilisin of claim 50 herein and to prepare a detergent composition comprising such a variant subtilisin and a surfactant of claims 53 and 54 herein. This is because Brode et al., Ballinger et al., and Gosselink et al. teach the advantages of introducing an amino acid substitution at the BPN'-correspondent position 62 in a subtilisin, such as the N62D substitution of Brode et al. where both of sub-groups I-S1 and I-S2 subtilases have asparagine present at the subtilisin BPN'-correspondent position 62, because Gosselink et al. teach that the prior art of subtilisin modification shows that a substitution of X76D, an N76D

substitution in the subtilisin BPN' amino acid sequence was well known to be advantageously combined with further amino acid substitutions in modifying a subtilisin to make it more effective for use in detergent compositions, and because Bott et al. teach that their paired, stabilizing, amino acid substitution including S87C provides improved thermal stability with autocatalytic cleavage characteristics nearly equivalent to the native subtilisins BPN'. It would also have been obvious to such an artisan at that time to prepare detergent compositions comprising such a variant sub-group I-S1 subtilisin as well as cellulases, lipases, and amylases, because Bott et al. teach that their paired, stabilizing, substitution with S87C provides improved thermal stability and autocatalytic cleavage characteristics nearly equivalent to that of the native subtilisin BPN and because Ballinger et al., Gosselink et al., and Brode et al. teach that it is advantageous to incorporate subtilisin variants having improved characteristics in detergent compositions. Such an artisan would have had a reasonable expectation of success in preparing a subtilisin variant that provides improved thermal stability and wash performance by combining the N62S/Q/D and N76D substitutions of Brode et al. with the stabilizing substitutions of Bott et al. that include S87C because the amino acid substitutions of Brode et al. occur at solvent-exposed positions of a subtilisin and the amino acid substitutions of Bott et al. provide enhanced thermal stability.

Conclusion

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

Any inquiry concerning this communication or earlier communications from the examiner should be directed to William W. Moore whose telephone number is 571.272.0933 and whose FAX number is 571.273.0933. The examiner can normally be reached Monday through Friday between 9:00AM and 5:30PM EST. If attempts to reach the examiner by telephone are unsuccessful, the examiner's Supervisory Primary Examiner, Dr. Kathleen Kerr Bragdon, can be reached at 571.272.0931. The official FAX number for all communications for the organization where this application or proceeding is assigned is 571.273.8300. Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is 571.272.1600.

William W. Moore
22 June 2007